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☐ 1: Hum Reprod 1994 Dec;9(12):2270-7

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Erratum in:

- Hum Reprod 1995 Apr;10(4):976

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### **CD56+ lymphoid cells in human first trimester pregnancy decidua as a source of novel transforming growth factor-beta 2-related immunosuppressive factors.**

**Clark DA, Vince G, Flanders KC, Hirte H, Starkey P.**

Department of Medicine, McMaster University, Hamilton, Ontario, Canada.

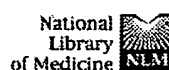
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The lymphomyeloid cells isolated from normal first trimester pregnancy decidua may be separated into a CD56+ population of natural killer (NK)-lineage cells with the morphology of granulated lymphocytes, and a CD56- population which includes other cell types. Unlike CD56+ NK cells in peripheral blood, decidual CD56+ cells lack type III Fc receptors (CD16) and did not express significant levels of either type I FcR (CD64) or type II FcR (CDw32). By contrast to the decidual CD56- cells, CD56+ cells could release biologically active transforming growth factor (TGF)-beta in vitro, detectable using an normal rat kidney fibroblast colony-forming assay. The CD56+ cells could be stained using an antibody specific for TGF-beta 2, and similarly staining cells could be detected in intact biopsies of normal pregnancy decidua. Bioactive TGF-beta is known to suppress the generation of cytotoxic cells in vitro, and high performance liquid chromatography fractionation of supernatants conditioned by CD56+ but not CD56- cells contained reproducible peaks of immunosuppressive activity at 40-45 and 15-20 kDa, similar to the TGF-beta 2 immunosuppressive activity in supernatants conditioned by unfractionated decidua.

PMID: 7536211 [PubMed - indexed for MEDLINE]

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☐ 1: Connect Tissue Res 1996;34(1):1-9

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## Engineering, expression and renaturation of targeted TGF-beta fusion proteins.

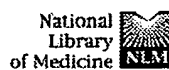
Tuan TL, Cheung DT, Wu LT, Yee A, Gabriel S, Han B, Morton L, Nimni ME, Hall FL.

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Research Institute Childrens Hospital Los Angeles, University of Southern California School of Medicine 90027, USA.

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This study reports the expression, purification, and renaturation of biologically active Transforming Growth Factor-beta 1 (TGF-beta 1) fusion proteins from *Escherichia coli* (*E. coli*). A prokaryotic expression vector was engineered to produce tripartite fusion proteins consisting of (i) a purification tag, (ii) a protease-sensitive linker/collagen binding domain, and (iii) a cDNA sequence encoding the active fragment of human TGF-beta 1. The expressed fusion proteins TGF-B1-F1 and TGF-B1-F2, located in inclusion bodies, were solubilized with 8 M urea and renatured using a glutathione redox-coupled system and protracted dialysis under several experimental conditions. The purification of the recombinant proteins was achieved by binding the His-tag of the fusion proteins on a Ni-NTA metal chelate column. The biological activity of the recombinant growth factor was demonstrated by its ability to inhibit mink lung (Mv1Lu) cell proliferation and/or to stimulate proliferation of NIH-3T3 mouse fibroblasts, where purified human platelet TGF-beta 1 served as a positive control. Purified TGF-B1-F1 and TGF-B1-F2 (collagen-binding) constructs exhibited anti-proliferative activities comparable to purified platelet TGF-beta 1, but at lower specific activities. Binding of the renatured TGF-B1-F2 fusion protein to collagen was demonstrated by stable binding on a collagen-conjugated Sephadex-G15 column. The high affinity binding was also demonstrated by the binding of 3H-collagen to the TGF-B1-F2 protein immobilized on a Ni-NTA column. The TGF-B1-F2 fusion protein bound to collagen coated surfaces with high affinity but exhibited comparatively lower biological activity than the fusion protein in solution, suggesting a potentially latent configuration. Taken together, these results demonstrate that biologically active TGF-beta 1 fusion proteins can be recovered from transformed bacteria by oxidative refolding; thus, providing a means for its high-yield production, purification, and renaturation from microorganisms. Furthermore, these results support the concept that auxiliary domains may be used to modulate and/or target TGF-beta 1 for specific applications.



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☐ 1: Nat Med 1995 Sep;1(9):932-7

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## Release and activation of platelet latent TGF-beta in blood clots during dissolution with plasmin.

Grainger DJ, Wakefield L, Bethell HW, Farndale RW, Metcalfe JC.

Department of Biochemistry, University of Cambridge, UK.

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Transforming growth factor beta 1 (TGF-beta 1) is a platelet-derived cytokine involved in both normal wound healing and scarring. We show that human platelets contain two pools of latent TGF-beta 1, which constitute more than 95% of the total TGF-beta assayed in whole platelets. During clotting, one pool, the large latent TGF-beta complex consisting of latent TGF-beta binding protein (LTBP), the latency-associated peptide (LAP) and the 25-kD mature TGF-beta 1 dimer is released into the serum. A second pool, which contains LAP but not LTBP, is retained in the clot, but can be released by RGD peptide. When the clot is dissolved by plasmin this bound TGF-beta 1 is gradually activated and released. If similar mechanisms operate in vivo, the clot will act as a slow-release capsule of TGF-beta 1 activity during wound healing.

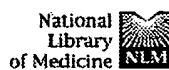
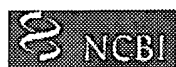
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PMID: 7585220 [PubMed - indexed for MEDLINE]

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☐ 1: Am J Reprod Immunol 1994 Mar-Apr;31(2-3):69-76 Related Articles, Books, LinkOut

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## Risk factors for antisperm antibodies in infertile men.

Heidenreich A, Bonfig R, Wilbert DM, Strohmaier WL, Engelmann UH.

Department of Urology, University of Cologne, Germany.

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**PROBLEM:** The prevalence of anti-sperm antibodies (ASAs) in the general population is 0 to 2%; the prevalence in infertile men is much higher at 7 to 26%. However, the role of ASAs in male infertility remains controversial to date. Although several risk factors for ASA development have been defined (such as testicular torsion, varicocele, cryptorchidism, vasectomy, and genital tract infection), there are no specific indications for ASA testing. **METHOD:** In order to examine if a single parameter exists identifying patients with elevated ASA titers, serum ASA testing was performed with an enzyme-linked immunosorbent assay (ELISA) in 226 consecutive male patients. The new assay, synchron ELISA (Synelisa) used in our study represents a new type of ELISA without fixation of the sperm surface antigens by formaldehyde or glutaraldehyde. Therefore, the quantitative assay is highly sensitive and reproducible since the structure of sperm surface antigens is not altered by the fixation process. **RESULTS:** The prevalence of ASAs in this population was 14%, while the prevalence of the control group was 2.5%. Of all factors analyzed only a history of vasectomy, an acute epididymitis, and an abnormal result in the bovine mucus penetration test was associated with elevated ASA titers ( $P < .001$ ). In addition, we could demonstrate a time related formation of ASAs in men after vasectomy. **CONCLUSIONS:**

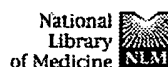
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☐ 1: J Cell Biol 1990 Apr;110(4):1361-7

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## Mechanism of activation of latent recombinant transforming growth factor beta 1 by plasmin.

Lyons RM, Gentry LE, Purchio AF, Moses HL.

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Department of Cell Biology, Vanderbilt School of Medicine, Nashville, Tennessee 37232.

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Medium conditioned by Chinese hamster ovary (CHO) cells transfected with the simian pre-pro-TGF beta 1 cDNA contains high levels of latent TGF beta 1. The amino-terminal region of the TGF beta 1 precursor is secreted and can be detected in the conditioned medium by immunoblotting using peptide antibodies specific for amino-terminal peptides. Chemical cross-linking of CHO-conditioned medium using bis-(sulfosuccinimidyl)-suberate (BS3) followed by immunoblot analyses indicates that latent recombinant TGF beta 1 contains both the cleaved amino-terminal glycopeptide and mature TGF beta 1 polypeptide in a noncovalent association and that this association confers latency. The data presented here do not support the involvement of a unique TGF beta binding protein(s) in latent recombinant TGF beta 1. Plasmin treatment of CHO-conditioned medium resulted in the appearance of TGF beta competing activity. In addition, immunoblot analysis of plasmin-treated CHO-conditioned medium indicates that the amino-terminal glycopeptide is partially degraded and that mature TGF beta 1 is released. Thus, activation of latent TGF beta 1 may occur by proteolytic nicking within the amino-terminal glycopeptide thereby causing a disruption of tertiary structure and noncovalent bonds, which results in the release of active, mature TGF beta 1. Acid activation of latent TGF beta, in comparison, appears to be due to dissociation of the amino-terminal glycopeptide from the mature polypeptide.

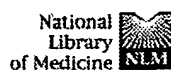
PMID: 2139036 [PubMed - indexed for MEDLINE]

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☐ 1: Am J Reprod Immunol 1995 Jul;34(1):52-64

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**A subset of patients with recurrent spontaneous abortion is deficient in transforming growth factor beta-2-producing "suppressor cells" in uterine tissue near the placental attachment site.**

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**Lea RG, Underwood J, Flanders KC, Hirte H, Banwatt D, Finotto S, Ohno I, Daya S, Harley C, Michel M, et al.**

Department of Biochemistry, McMaster University, Hamilton, Ontario, Canada.

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**PROBLEM:** To determine if patients with unexplained recurrent miscarriage have a deficiency of decidual immunosuppressor cells that produce transforming growth factor beta type 2, as has been found in mice with abortion due to rejection and/or trophoblast failure. **METHODS:** Decidual biopsy specimens were taken as near to the placental attachment site as possible under ultrasound guidance from first trimester legal termination (control) patients with recurrent miscarriage and non-viable pregnancy, and from patients with sporadic missed abortion. The tissue was tested for TGF beta-2+ suppressor cells by in situ hybridization, immunohistochemistry, and analysis of supernatants. **RESULTS:** TGF beta-2-related suppressor molecules similar but not identical to those identified in pregnant mice were released by decidual lymphoid cells. Fifty percent of 14 recurrent miscarriage patients showed a lack of suppressor cells and 59% were subnormal in comparison to 20 controls and 5 sporadic miscarriage patients, where 80-85% of the patients had detectable suppressor cells. **CONCLUSIONS:** Suppressor cell deficiency is compatible with a role for rejection and/or trophoblast failure in some patients with recurrent miscarriage. Presence of suppressor cells in most patients with missed abortion (4/5) is compatible with an alternative cause of fetal death, similar to findings reported in genetic fetal death mice.

PMID: 7576131 [PubMed - indexed for MEDLINE]

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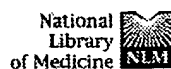
on the presence of soluble signals from fetal trophoblast may be explainable by the ability of cells bearing TcR gamma delta to recognize and react to placental trophoblast cell antigen.

PMID: 9160738 [PubMed - indexed for MEDLINE]

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☐ 1: J Clin Immunol 2000 Nov;20(6):453-7

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## Elevation of transforming growth factor-beta1 is associated with recurrent miscarriage.

Ogasawara MS, Aoki K, Aoyama T, Katano K, Iinuma Y, Ozaki Y, Suzumori K.

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Department of Obstetrics and Gynecology, Nagoya City University Medical School, Nagoya, Japan.

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To investigate the significance of transforming growth factor-beta1 (TGF-beta1) in reproduction we have compared plasma levels in normal pregnant women and patients suffering miscarriages. We examined 188 normal pregnant women and 12 pregnant women with miscarriages. Eight women with severe recurrent miscarriages (mean +/- SD of previous number of miscarriages; 10.4 +/- 2.4 times) were also examined before conception; 34 nonpregnant women served as controls. Plasma TGF-beta1 level increased with the gestational week and returned within the normal range 1 month after delivery. The levels among pregnant women with miscarriages (mean +/- SD; 2.44 +/- 0.83 ng/ml) were significantly higher than those of pregnant controls (1.74 +/- 0.95 ng/ml) of matched gestational weeks; levels among nonpregnant women with severe recurrent miscarriages were extremely elevated (4.1 +/- 3.04 ng/ml) compared to the control value (1.34 +/- 0.59 ng/ml). These data suggest that TGF-beta1 may be necessary to maintain pregnancy but also may be a risk factor for recurrent miscarriages.

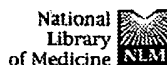
PMID: 11202235 [PubMed - indexed for MEDLINE]

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☐ 1: Am J Reprod Immunol 2000 Sep;44(3):129-35

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## Is the paternal mononuclear cells' immunization a successful treatment for recurrent spontaneous abortion?

Ramhorst R, Agriello E, Zittermann S, Pando M, Larriba J, Irigoyen M, Cortelezzi M, Auge L, Lombardi E, Etchepareborda JJ, Contreras Ortiz C, Fainboim L.

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Division Immunogenetica, Hospital de Clinicas, Facultad de Medicina, UBA, Buenos Aires, Argentina.

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**PROBLEM:** Alloimmunization as a treatment for recurrent spontaneous abortion (RSA) is still controversial due to the lack of enough controls to evaluate its effectiveness. The present study was conducted to compare the live birth rate in the presence or absence of immunotherapy. **METHOD OF STUDY:** Ninety-two women with RSA (79 primary [PA] and 13 secondary aborters[SA]) received immunotherapy. Thirty-seven RSA couples not receiving paternal alloimmunization, constituted the "control" group. **RESULTS:** The pregnancy rate in alloimmunized was 58 vs 46% in the control group. The live birth increased from 71% in the controls to 88% after immunotherapy. The alloimmunization induced mixed lymphocyte reaction blocking factors (MLR BF<sub>s</sub>) in 79% of women. However, they were also present in 83% of immunized women experiencing a new abortion. **CONCLUSION:** These results indicate that alloimmunization may be useful in the treatment of RSA.

Publication Types:

- Clinical Trial

PMID: 11028898 [PubMed - indexed for MEDLINE]

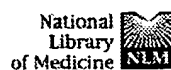
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☐ 1: Am J Reprod Immunol 2000 Nov;44(5):289-92

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## Adverse influence of numbers of previous miscarriages on results of paternal lymphocyte immunization in patients with recurrent spontaneous abortions.

Katano K, Aoki K, Ogasawara MS, Suzumori K.

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Department of Obstetrics and Gynecology, Nagoya City University Medical School, Nagoya, Japan.

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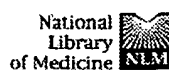
**PROBLEM:** To determine whether an increase in the number of previous miscarriages in recurrent spontaneous abortion patients is a risk factor in subsequent pregnancies with paternal lymphocyte immunotherapy. **METHOD OF STUDY:** Live birth rates with reference to previous abortion numbers in recurrent spontaneous abortion patients were statistically compared between paternal lymphocyte immunotherapy and control groups, the latter retrospectively researched using historical data before 1981 in our clinic. **RESULTS:** The overall live birth rate was 73% (169/232) in the immunotherapy group, and 48% (47/97) in controls ( $P < 0.05$ ). According to previous abortion numbers, the rates were 77% (114/148) versus 55% (36/65) ( $P < 0.05$ ) for three previous abortions, 70% (40/57) versus 38% (8/21) ( $P < 0.05$ ) for four and 56% (15/27) versus 27% (3/11) (not significant) for five, in the study and control groups, respectively. **CONCLUSIONS:** The results confirm the efficacy of paternal lymphocyte immunotherapy, but demonstrate that the success rate deteriorates with the number of previous miscarriages.

PMID: 11125791 [PubMed - indexed for MEDLINE]

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☐ 1: J Clin Invest 1990 Dec;86(6):1976-84

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**Recombinant latent transforming growth factor beta 1 has a longer plasma half-life in rats than active transforming growth factor beta 1, and a different tissue distribution.**

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**Wakefield LM, Winokur TS, Hollands RS, Christopherson K, Levinson AD, Sporn MB.**

Laboratory of Chemoprevention, National Cancer Institute, Bethesda, Maryland 20892.

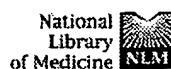
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Transforming growth factor beta 1 (TGF-beta 1) is a key regulator of cell growth and differentiation. Under normal physiological conditions, it is made as a biologically latent complex whose significance is unknown. Previous work has indicated that active TGF-beta 1 has a very short plasma half-life in rats (Coffey, R. J., L. J. Kost, R. M. Lyons, H. L. Moses, and N. F. La-Russo. 1987. J. Clin. Invest. 80:750-757). We have investigated the possibility that latent complex formation may extend the plasma half-life of TGF-beta 1 and alter its organ distribution. Radiolabeled latent TGF-beta 1 was formed by noncovalent association of 125I-TGF-beta 1 with the TGF-beta 1 precursor "pro" region from recombinant sources. TGF-beta 1 in this latent complex had a greatly extended plasma half-life (greater than 100 min) in rats compared with active TGF-beta 1 (2-3 min). Whereas active TGF-beta 1 was rapidly taken up by the liver, kidneys, lungs, and spleen and degraded, TGF-beta 1 in the latent complex was largely confined to the circulation, and was less than 5% degraded after 90 min. The pharmacokinetics of TGF-beta 1 in the latent complex were shown to be critically dependent on the degree of sialylation of the complex. The results suggest that formation of latent complexes may switch endogenous TGF-beta 1 from an autocrine/paracrine mode of action to a more endocrine mode involving target organs distant from the site of synthesis.

PMID: 2254455 [PubMed - indexed for MEDLINE]

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☐ 1: J Cell Physiol 1989 Oct;141(1):170-80

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## Modulation of TGF-beta type 1 receptor: flow cytometric detection with biotinylated TGF-beta.

Newman W, Beall LD, Bertolini DR, Cone JL.

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Department of Immunology, Otsuka Pharmaceutical Co., Rockville, Maryland 20850.

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Transforming growth factor beta type 1 (TGF-beta 1) was reacted with NHS-biotin to yield a derivative of TGF-beta 1 which was biotinylated on lysine residues. The biotinylated form of TGF-beta 1 was separated from the unreacted material by reverse phase chromatography. In three separate bioassays, the derivatized peptide was as active as the starting material. The use of FITC-avidin in conjunction with flow cytometry demonstrated that the binding of biotinylated TGF-beta 1 to its receptor is saturable, competable, and specific. A 100-fold molar excess of underivatized TGF-beta 1 gave 85% inhibition of binding of the biotinylated peptide to the mink lung cell line CCL-64, while TGF-beta 2 showed no inhibition of binding, nor did insulin, calcitonin, or TGF-alpha. Both CCL-64 cells and human umbilical vein endothelial cells showed a density-dependent down-regulation of receptor expression in culture. Several factors were examined that might mediate this effect. The down-regulation was shown not to be due to the secretion of an active form of TGF-beta 1. The extracellular matrix from high-density cells did not decrease expression of the receptor. Fibronectin, collagen, and gelatin were also unable to signal changes in receptor expression, even though in other systems such matrix components can regulate the responsiveness of cells to TGF-beta 1. Lastly, staining simultaneously for DNA content and TGF-beta 1 receptor expression showed that there was no correlation between cell cycle and receptor levels.

PMID: 2550480 [PubMed - indexed for MEDLINE]

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1: Biol Reprod 1996 Jul;55(1):54-61

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## Characterization of human sperm antigens reacting with sperm antibodies from autologous serum and seminal plasma in an infertile population.

**Paradisi R, Bellavia E, Pession A, Venturoli S, Flamigni C.**PubMed  
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Department of Obstetrics, Gynecology and Reproductive Biology, S. Orsola Hospital, Bologna, Italy.

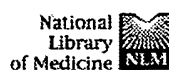
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Immunoblotting techniques were used to characterize the reactivity of human sperm antigens with sperm antibodies from an infertile population. Sperm antigens of each individual were tested with autologous sperm antibodies present in serum and seminal plasma in order to construct a preliminary map of the antigens of the infertile spermatozoon and to compare the qualitative differences in the antigenic profile between fertile and infertile subjects. A total of 61 infertile males, comprising 51 subjects having poor semen quality and 10 subjects with no abnormalities in semen analysis, entered the study; 55 subjects with proven fertility served as controls. Infertile subjects often showed specific immunoreactivity to 50-, 55-, 57-, 62-, and 72-kDa proteins in serum and to 57- and 62-kDa proteins in seminal plasma. As to comparison of immunoreactivities between fertile and infertile individuals, the sperm antigens may be divided into three groups. Group 1 antigens (50-, 69-, and 72-kDa proteins) were recognized by sperm antibodies present in both populations; group 2 antigens (57- and 62-kDa proteins), by sperm antibodies typical of the infertile population; group 3 antigens (45-, 55-, and 85-kDa proteins), by sperm antibodies typical of the fertile population. This classification shows that the infertile spermatozoon differs substantially at the immunogenic level from the fertile spermatozoon. The group 2 antigens seem to be involved in a relevant step in the reproductive process and hence have been termed "fertility-related antigens."

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Identification of human sperm surface glycoproteins recognized by autoantisera from immune infertile men, women, and vasectomized men.  
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J Reprod Immunol. 2002 Jan;53(1-2):1-12.

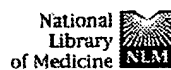
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## Risk factors for antisperm antibodies in infertile men.

Heidenreich A, Bonfig R, Wilbert DM, Strohmaier WL, Engelmann UH.

Department of Urology, University of Cologne, Germany.

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**PROBLEM:** The prevalence of anti-sperm antibodies (ASAs) in the general population is 0 to 2%; the prevalence in infertile men is much higher at 7 to 26%. However, the role of ASAs in male infertility remains controversial to date. Although several risk factors for ASA development have been defined (such as testicular torsion, varicocele, cryptorchidism, vasectomy, and genital tract infection), there are no specific indications for ASA testing. **METHOD:** In order to examine if a single parameter exists identifying patients with elevated ASA titers, serum ASA testing was performed with an enzyme-linked immunosorbent assay (ELISA) in 226 consecutive male patients. The new assay, synchron ELISA (Synelisa) used in our study represents a new type of ELISA without fixation of the sperm surface antigens by formaldehyde or glutaraldehyde. Therefore, the quantitative assay is highly sensitive and reproducible since the structure of sperm surface antigens is not altered by the fixation process. **RESULTS:** The prevalence of ASAs in this population was 14%, while the prevalence of the control group was 2.5%. Of all factors analyzed only a history of vasectomy, an acute epididymitis, and an abnormal result in the bovine mucus penetration test was associated with elevated ASA titers ( $P < .001$ ). In addition, we could demonstrate a time related formation of ASAs in men after vasectomy. **CONCLUSIONS:**

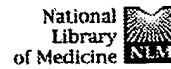
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## Stress-triggered abortion: inhibition of protective suppression and promotion of tumor necrosis factor-alpha (TNF-alpha) release as a mechanism triggering resorptions in mice.

Arck PC, Merali FS, Manuel J, Chaouat G, Clark DA.

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Department of Medicine, McMaster University, Hamilton, Ontario, Canada.

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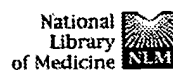
**PROBLEM:** Stress adversely affects pregnancy outcome and has been implicated as an abortogen in both animals and humans. However, the mechanisms whereby stress abortions are largely unknown. Alloimmunization can prevent stress-triggered abortion, and immunization is known to increase transforming growth factor-beta 2 (TGF-beta 2)-related suppressive activity. **METHOD:** To investigate these mechanisms, DBA/2J males were mated to CBA/J or C3H/HeJ females, and the pregnant females were exposed to ultrasonic sound stress for a period of 24 h between day 4.5 to 8.5 of pregnancy. **RESULTS:** Ultrasonic stress significantly elevated the resorption rate with a peak effect on day 5.5 in the CBA/J females and on day 4.5 in the LPS-resistant C3H/HeJ females. The tumor necrosis factor-alpha (TNF-alpha) release from the decidua was also elevated and the TGF-beta 2-mediated suppressive activity was significantly decreased. The resorption rate only increased when the TNF-alpha/TGF-beta 2 ratio was increased compared to the control. **CONCLUSION:** These data suggest that stress may inhibit protective suppressor mechanisms and promote secretion of abortogenic cytokines such as TNF-alpha. Possible mechanisms are discussed.

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**Release of a transforming growth factor (TGF)-beta 2-related suppressor factor from postimplantation murine decidual tissue can be correlated with the detection of a subpopulation of cells containing RNA for TGF-beta 2.**

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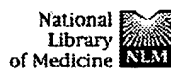
**Lea RG, Flanders KC, Harley CB, Manuel J, Banwatt D, Clark DA.**

Department of Medicine, McMaster University, Hamilton, Ontario, Canada.

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Postimplantation murine decidual tissue from allopregnant C3H mice has been shown to release in vitro a potent immunosuppressive factor closely related to transforming growth factor (TGF)-beta 2 but slightly lower in apparent molecular weight. Decidual suppressor factor (DSF) activity was first detected in decidual tissue supernatant at day 9.5 of gestation and reached a plateau by day 10.5 to 12.5. By Northern analysis of decidual and placental tissue with a simian TGF-beta 2 probe, two characteristic TGF-beta 2 mRNA transcripts were detected in decidual tissue. In situ hybridization analysis of C3H implant sites, with the simian (pGEM-G1G2) TGF-beta 2 riboprobe, revealed a small population of TGF-beta 2+ cells localized to postimplantation decidua basalis and metrial gland cell area after day 8.5. On and before day 8.5, when DSF was not detectable, few TGF-beta 2 mRNA+ cells were detected. To test for TGF-beta release in situ, sections of uterine tissue were stained with antibody specific for TGF-beta 2, that identified DSF in Western blots. In postimplantation tissues (day 9.5, 12.5) patchy anti-TGF-beta 2 staining was seen over decidual tissue. Before day 9.5, slight and diffuse staining over decidual tissue was present with more marked staining of extradecidual tissue. Very little staining was noted over day 9.5 decidual tissue by using anti-TGF-beta 1 antibody as a control; however, some staining was seen over postimplantation fetal trophoblast and myometrial tissue. Fractionation of disaggregated postimplantation decidua by velocity sedimentation revealed that TGF-beta 2 mRNA+ cells were predominantly small and sedimented in the same fraction(s) as those cells previously shown to release DSF in vitro. Thus, the release of TGF-beta 2 related DSF correlates with the in situ detection of TGF-beta 2 mRNA and the in situ release of TGF-beta 2 peptide. These studies suggest that DSF may be a form of TGF-beta 2 released by a population of small lymphocytic decidual suppressor cells.

PMID: 1730871 [PubMed - indexed for MEDLINE]



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**Immunotherapy before and during pregnancy improves pregnancy outcome in women who suffer from recurrent abortion and did not benefit from immunotherapy before pregnancy.**

**Maejima M, Fujii T, Yamashita T, Hara N, Hamai Y, Miki A, Kozuma S, Okai T, Shibata Y, Taketani Y.**

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Department of Obstetrics and Gynecology, Faculty of Medicine, University of Tokyo, Japan.

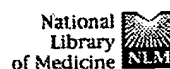
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**PROBLEM:** The appropriate modality of immunotherapy with the husband's mononuclear cells in women with a history of recurrent abortion who aborted despite the immunotherapy performed before pregnancy was explored. **METHOD OF STUDY:** Nineteen patients who had suffered from recurrent abortion who had received the immunotherapy only before pregnancy and had aborted were treated with further immunotherapy performed either only before pregnancy or twice: before and during pregnancy. **RESULTS:** In 9 out of the 19 women who received further immunotherapy before pregnancy, 2 had healthy babies and 7 aborted again. In the remaining 10 patients who received further immunotherapy twice, before and during pregnancy, 8 had healthy babies and 2 aborted again. **CONCLUSION:** Our results indicate that immunotherapy performed before and during pregnancy produces a better outcome compared with that performed only before pregnancy, especially in patients who showed no benefit from the immunotherapy performed only before pregnancy.

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**Seminal transforming growth factor beta1 stimulates granulocyte-macrophage colony-stimulating factor production and inflammatory cell recruitment in the murine uterus.**

**Tremellen KP, Seamark RF, Robertson SA.**

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Department of Obstetrics and Gynaecology, University of Adelaide, South Australia, Australia.

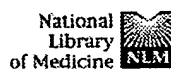
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Mating in rodents evokes an inflammatory-like reaction within the uterine endometrium, characterized by extensive infiltration and activation of macrophages, dendritic cells, and granulocytes. This response is initiated when seminal vesicle gland-derived factors in the ejaculate stimulate uterine epithelial cells to release proinflammatory cytokines including granulocyte-macrophage colony-stimulating factor (GM-CSF). Experiments in which seminal vesicle secretions were fractionated by Sephacryl S-400 chromatography and assayed in vitro for GM-CSF-stimulating activity revealed that the seminal moiety coeluted with transforming growth factor beta1 (TGFbeta1) in the 150-440-kDa range and was neutralized by anti-TGFbeta1 antibodies. Comparable amounts of recombinant TGFbeta1 stimulated GM-CSF release in cultures of uterine epithelial cells from estrous mice and, when instilled into the uterine lumen, caused an increase in GM-CSF content and an infiltration of leukocytes into the endometrium similar to the postmating response. These results show that seminal vesicular fluid contains TGFbeta1 at levels sufficient to be the primary causative agent in the postmating inflammatory cascade through induction of GM-CSF synthesis by uterine epithelial cells. Seminal TGFbeta1 is thus implicated as a key factor in initiation of the remodeling events and immunological changes that occur in the uterus during the preimplantation period of pregnancy.

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**Decidua-associated suppressor cells in abortion-prone DBA/2-mated CBA/J mice that release bioactive transforming growth factor beta2-related immunosuppressive molecules express a bone marrow-derived natural suppressor cell marker and gamma delta T-cell receptor.**

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Clark DA, Merali FS, Hoskin DW, Steel-Norwood D, Arck PC, Croitoru K, Murgita RA, Hirte H.

Department of Medicine, McMaster University, Hamilton, Ontario, Canada.  
clarkd@fhs.csu.mcmaster.ca

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The decidua of allopregnant mice contains a novel population of Thy1 Lyt1 CD4 CD8 asialoGM1- non-B small lymphocytic suppressor cells that release transforming growth factor (TGF) SS2-related suppressor molecules. The "null" phenotype of this cell population is similar to some bone marrow-derived natural suppressor cell (NSC) populations, and the latter may release TGF(beta)s. We now report that the TGF beta2-producing suppressor cells in the uterine decidua of DBA/2-mated CBA/J female mice-linked to prevention of abortions-are inactivated effectively by 1E5/B5.1 but not by 2C1.1 rat monoclonal antibodies to murine pregnancy-associated splenic NSC in the presence of complement. Immunostaining of a subpopulation of cells in decidua with 1E5/B5.1 but not with 2C1.1 was shown by flow cytometry. Release of suppressor factor was also abrogated by 1E5/B5.1 + complement but not by 2C1.1 + complement, and the suppressor factor was specifically neutralized by anti-TGF beta2 and not by anti-TGF beta3. Splenic pregnancy NSC are susceptible to 2C1.1, produce TGF beta1, and express CD3 and alpha beta T-cell receptor (TcR) chains. Release of suppressor factor by the decidual NSC was abrogated by treatment with anti-CD3 (145 2C11) and anti-TcR gamma delta (GL4) monoclonal antibodies + complement, but not by anti-TcR alpha beta (H57) + complement; and cells sorted using anti-TcR gamma delta (GL3) released suppressive activity in vitro. Slightly more suppressive activity was released by implantation-site decidua where there was no epithelium than from epithelialized inter-implantation-site decidua; no significant activity was released from placental tissue, but combining implantation-site tissue with placental tissue led to release of enhanced levels of immunosuppressive activity. There appear to be subtypes of bone marrow-derived TcR+ NSC with different phenotypes and tissue localization patterns in pregnancy. The previously reported dependence of decidual NSC activity